

## Article

### Seasonal activity and spatial distribution pattern of *Eotetranychus frosti* (Acari: Tetranychidae) in an unsprayed apple orchard of Kermanshah, Western Iran

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#### Abstract

Population density and spatial distribution pattern of *Eotetranychus frosti* (McGregor) was determined in an unsprayed apple orchard of Kermanshah, Western Iran, from 31<sup>st</sup> May till 7<sup>th</sup> November 2007. On each sample date 130 leaves were selected, with a relative variation of 4.31%. Different methods of indices of dispersion, regression models (Taylor and Iwao), Morisita's index and Lloyd's mean crowding to mean was used to determine the spatial distribution pattern of this mite. The mean population density of this mite per leaf was 2.079. The highest population density of *E. frosti* was in 9<sup>th</sup> August (4.938 per leaf). Index of dispersion ( $S^2/m= 6.516$ ), aggregation index ( $1/k= 2.653$ ) and Lloyd's mean crowding to mean ( $m^*/m=3.653$ ) showed an aggregated distribution for this species. The Taylor's power law and Iwao's patchiness regression methods showed aggregated spatial distribution too. Some changes in spatial distribution pattern from aggregated to random were observed using Morisita's index in different sampling dates. Three different coefficients of Taylor, Iwao and K were used to optimize the sample size for future studies. The lowest optimized sample size (34.70) was obtained by using the Taylor's coefficients. Spatial distribution parameters of *E. frosti* can be used to improve a sampling program of this mite in a framework of an integrated pest management system.

**Keywords:** *Eotetranychus frosti*, population density, spatial distribution, optimum sample size

#### Introduction

High population levels of cosmopolitan mites of the family Tetranychidae can be seen as pests in some perennial agro-ecosystems like apple orchards (Kasap 2005). They ingest leaf cell contents, thus reducing plant photosynthesis, potentially decreasing fruit quality (Prischmann *et al.* 2005). Using acaricides on calendar-based programs, as currently

being used as a control method for these pests (Greco *et al.* 2005), has resulted in pest resistance and residue on products.

Orchard management practices usually cause outbreaks of spider mites followed by disrupted population of natural enemy or induce mite migration from the ground cover into trees (Alston 1994). Biological control is a useful alternative to pesticides for managing various arthropod pests and the predator mites are the most important factors in reducing tetranychids and other plant feeding mite populations (Gerson *et al.* 2003).

To evaluate natural enemies' potential to reduce their prey density and increase system persistence, having information about spatial distribution of prey is critical (Slone and Croft 1998). It can also be used to investigate population dispersion behavior, establish a precise sampling plan and program sequential sampling (Margolis *et al.* 1984 ; Deligeorgidis *et al.* 2002), detect pest levels for justifying control measures and assess crop loss (Kavallieratos *et al.* 2005).

*Eotetranychus frosti* (McGregor) is one of the most important phytopagous tetranychid mites in Kermanshah orchards and have been reported on apples, figs and plums in different parts of province (Darbemamieh 2008). This mite has been reported from orchards in other parts of Iran (Khodayari *et al.* 2010, Kamali *et al.* 2000) and is an injurious pest in apple orchards.

Using information of sample mean, variance and size, the variance-mean relationships of Taylor (1961) and Iwao (1968) have been effectively used in many sampling procedures (Beers and Jones 2004; Hamilton and Hepworth 2004). Quantitative knowledge of spatial distribution patterns of plant feeding mites is essential to develop reliable sampling plans for monitoring pest abundance (Onzo *et al.* 2005; Athanassiou *et al.* 2005). Finding out the generic coefficients eliminate experimental needs for large sample size (Ifoulis and Savopoulou-Soultani 2006).

The aim of this study is monitoring the seasonal activity of *E. frosti*, its spatial distribution in Kermanshah natural conditions and finding primitive information about its natural enemies and control to establish a biological control for this mite on apples. The calculated common coefficients of this study could be used in various apple cultivars and also some similar orchards.

## **Material and methods**

### *Sampling Procedure*

In this study, one apple leaf was selected as a sample unit. Leaves were selected randomly and from all parts of canopy avoiding biased estimate of population mean. Samples were taken at 9-12 in the morning from May 31<sup>st</sup> till November 7<sup>th</sup> 2007 by 10-day intervals. The number of mobile stages of *E. frosti* per leaf was counted at the same day using stereomicroscope (Olympus). After primary sampling, the relative variation (*RV*) was calculated according to Hillhouse and Pitre (1974) to evaluate the accuracy of data.

### *Spatial Distribution*

The spatial distribution of *E. frosti* was determined by the following five methods: index of dispersion, Morisita's coefficient of dispersion, Lloyd's mean crowding and

regression techniques including Taylor's Power Law and Iwao's Patchiness (Taylor 1961, Lloyd 1967, Iwao 1968, Darbemamieh *et al.* 2009).

#### Optimum Number of Sample Units

The optimum sample size, smallest number of sample units with precision of estimates is calculated by using coefficients  $a$  and  $b$  within Taylor's power law in equation 1:

$$1) \quad N_{opt} = a \left( \frac{t_{\alpha/2}}{D} \right)^2 (\mu^{b-2})$$

Where  $N_{opt}$  = optimum sample size,  $t_{\alpha/2}$  = t- student of table,  $\mu$  = mean density,  $a$  and  $b$  = Taylor's coefficients and  $D$  = the range of accuracy.

The optimum sample size is derived from equation 2:

$$2) \quad N_{opt} = \left( \frac{t_{\alpha/2}}{D} \right)^2 \left( \frac{1}{\mu} + \frac{1}{k} \right)$$

using  $k$  in negative binomial distribution equation 3:

$$3) \quad \frac{1}{k} = \frac{\sigma^2 - \mu}{\mu^2}$$

This estimation can also be done by Iwao's patchiness regression method coefficients ( $\alpha$  and  $\beta$ ) in formula 4 (Wilson 1985):

$$4) \quad N_{opt} = \left( \frac{t_{\alpha/2}}{D} \right)^2 \left( \frac{\alpha + 1}{\mu} + (\beta - 1) \right)$$

## Results

Biggest calculated  $RV$  and reliable sample size were 4.31% and 130, respectively. Population fluctuations of *E. frosti* (Figure 1) observed from May 31<sup>st</sup> until November 7<sup>th</sup>. First observation of *E. frosti* was made by the end of May but its population didn't increase till June 20<sup>th</sup>, peaking on August 9<sup>th</sup>, 2007 (4.938 per leaf). Sharp decline in population density was seen at the end of the September and continued by the end of the monitoring period.

**Table 1.** Estimated values of intercept and slope for *Eotetranychus frosti* in 2007 by regression analysis of Taylor's Power Law and Iwao's Patchiness Regression

Method	$a$	$b$	$r^2$	$P$ - value
Taylor	0.434	1.277	0.944	<0.0001
Iwao	0.146	2.315	0.798	<0.0001

Both regression methods fit the data well for examined species (Table 1). The index of dispersion ( $I_D$ ) and the  $m^*/m$  values for *E. frosti* were significantly greater than 1 (Table 3), which means this species exhibited aggregated behavior in its habitat. The  $1/k$  values calculated suggests aggregated pattern of dispersion for this mite too (Table 2). There were some differences in Morisita's index values of species but in most sampling dates, the index was significantly greater than 1.96 (Table 3), suggesting that the spatial distribution of this tetranychid mite was aggregated.

**Table 2.** Estimated parameters by Lloyd mean crowding, index of dispersion, Lloyd mean crowding to mean and common  $k$  for *Eotetranychus frosti* in 2007

Species	$m$	$S^2$	$1/k$	$S^2/m$	$m^*$	$m^*/m$	$Id$	$Z$
<i>E. frosti</i>	2.079	13.547	2.653	6.516	7.595	3.653	9317.663	83.051

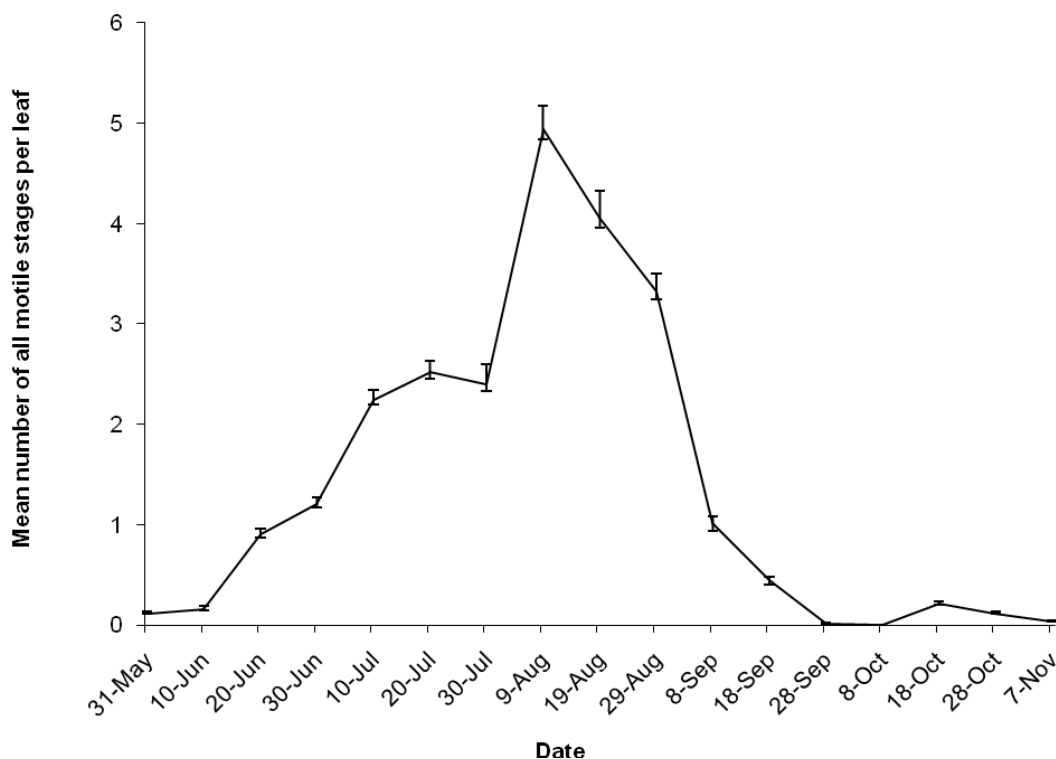
In this study, absolute counts of *E. frosti* motile stages were used to develop generic coefficients of regression techniques based on large amount of data. The sample size was re-calculated using  $k$  in negative binomial distribution and Taylor's and Iwao's coefficient ( $a$ ,  $b$ ,  $\alpha$  and  $\beta$ ). Lowest estimate of sample size calculated with Taylor's equation (34.70 leaves per sampling date) and Highest calculated with K (425.37 leaves per sampling date). This estimation with Iwao's equation was 318.57 leaves per sampling date. For acquiring greater precision, we adopted the 20% level, whereas in IPM programs, 25 or 30% level is acceptable.

**Table 3.** Morisita's index and  $Z$  values for *Eotetranychus frosti* in different sampling dates of 2007

Date	<i>E. frosti</i>	
	$I_{\delta}$	$Z$
31 May	11.143	708.711
10 Jun	9.286	413.533
20 Jun	2.069	9.576
30 Jun	1.763	5.129
10 Jul	1.459	1.649
20 Jul	1.810	2.595
30 Jul	4.035	10.197
09 Aug	2.013	1.654
19 Aug	3.150	4.277
29 Aug	2.182	2.869
08 Sep	2.315	10.445
18 Sep	2.831	33.091
28 Sep	0.000	-524.047
08 Oct	-	-
18 Oct	7.778	263.102
28 Oct	3.714	198.655
07 Nov	13.000	2515.424

## Discussion

Highest population density occurred on August with peak amount of 4.938 mites per leaf on August 9<sup>th</sup>. Khodayari *et al.* (2010) observed this peak in mid-September with much lower population density (2.46 mites per leaf). The population density of *E. frosti* increased in August as a result of increase in temperature and dryness. Furthermore, the sharp decline in *E. frosti* population that was observed from September 8<sup>th</sup> may have been due to weather or the predator's activity (Figure 1). Some predators like *Zetzellia mali* (Ewing) (Acari: Stigmaeidae) and *Typhlodromus bagdasarjani* (Wainstein & Arutunj) (Acari: Phytoseiidae) have been collected from its natural environment in sampling place (Darbemamieh 2008; Darbemamieh *et al.* 2009; Darbemamieh *et al.* 2011). Khodayari *et al.* (2010) reported *Z. mali* as an active predator of *E. frosti* in apple orchards of Maragheh, northwestern of Iran. Lower densities of *E. frosti* in Maragheh are related to high population density of *Z. mali* that can feeds on eggs and immature stages of *E. frosti* and some other mites like *Tydeus longisetosus* (ElBagouy and Momen)(Acari: Tydeidae) .



**Figure 1.** Population fluctuation of *Eotetranychus frosti* on apple leaves in 2007

The results of Taylor and Iwao regression methods showed that the spatial distribution pattern of this spider mite was aggregated. Data demonstrated better fitness to Taylor's ( $r^2 = 0.944$ ) for *E. frosti* when compared to Iwao's model ( $r^2 = 0.798$ ) but have good fitness to the both. Taylor's power law as well as Iowa's patchiness regression has been widely used for dispersion evaluating, data normalizing for statistical analysis and sampling protocols for many insects (Deligeorgidis *et al.* 2002). Random distribution for this mite was reported by Khodayari *et al.* (2010) using regression methods of Taylor's power law and Iowa's patchiness regression that can be obtained as a result of lower sample size (61.66 leaf per sampling date) and accuracy ( $RV$ : 6).

Observed aggregation in spatial distribution for this mite with the index of dispersion, Lloyd mean crowding and  $1/k$  values suggests that presence of an individual mite at one point may cause increased probability of another individual being nearby. Using index of dispersion ( $I_D$ ) and the Lloyd's mean crowding values aggregation distribution was reported by Khodayari *et al.* (2010) for this mite too. Aggregated distribution of spider mites has been shown in other studies too (Nuchman 1984; Strong *et al.* 1997). It seems that dispersion indexes are convenient decision making factors for management programs because of their fast way of calculating and simple results.

Being greater than 1.96 for Morisita's index values in most sampling dates, suggests that the spatial distribution of this species was aggregated. Some changes in distribution pattern observed by this method are results of different stages and conditions in each sampling date. Since Morisita's coefficient estimates spatial distribution using the mean and variance of each sampling date separately, this index is more accurate than the dispersion index. Showing one distribution per each date can be used to understand details of dispersion in different sampling dates that would be useful for research strategies more than management programs. Changes in distribution of *E. frosti* in two dates in July and August from aggregated to random can be partly due to increase in population density or movement of nymphs from clumped egg locations. It seems that distribution pattern in most of sampling dates could be used as a base for management decisions. Uniform distribution observed in late September was a result of low population density (Darbemamieh *et al.* 2011).

The spatial distribution of population individuals in an ecosystem can be a result of behavioral patterns and environment. Despite of parameters such as rate of population increase and reproduction that will change from one generation to another, spatial distribution is partially constant and is a character of species (Taylor 1984). Clump laying behavior and slow movement of *E. frosti* could be accounted as possible reason for their aggregated spatial distribution. Furthermore, the tolerance of a species to the environmental factors like temperature, relative humidity and low food density which in turn refers to population genetic can determine the spatial distribution being used as a distinguishing factor of adjacent species. Spatial distribution of prey can determine its natural enemy distribution, especially the predators' one. For example searching rate of phytoseiids in aggregated populations of spider mites is higher than that observed in tetranychid populations with random distribution (Kim & Lee 1993).

Lowest estimate of sample size which was calculated with Taylor's equation for *E. frosti* demonstrated better fitness of this model for evaluating sample size to this mite. In comparative studies of models, Taylor's power law model was usually showed to fit

the spatial dispersion better than Iwao's model to achieve a desired precision of estimates (Afshari *et al.* 2009; Darbemamieh *et al.* 2009). Basically, Taylor's method results in almost half the necessary sample size compared to common  $k$  or Iwao's method. Iwao's method was originally derived with close reference to theoretical distribution models (Davis 1994). This may account for the observed similarity with the calculated amounts using the common  $k$  and Iwao's method. In contrast, as a purely empirical model, Taylor's power law doesn't have such definite theoretical basis (Kuno 1991). However, Taylor's power law has been widely used because of its statistical stability. In this study for acquiring greater precision, we adopted the 20% level, whereas in IPM programs, 25 or 30% level is acceptable. Therefore the optimum sample size calculated at the beginning of work was suitable for this species.

Seasonal activity and spatial distribution parameters of the *E. frosti* can be used as a foundation for sampling programs and can also be used in estimating this mites' population density which in turn could be used in classical control strategies or integrated pest management programs for apple orchards.

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## چکیده

تراکم جمعیت و الگوی توزیع فضایی کنه (*Eotetranychus frosti* (McGregor)) از ۸۶/۳/۱۱ تا ۸۶/۸/۱۶ در یک باغ سیب سمپاشی نشده در شهرستان کرمانشاه مطالعه شد. تعداد ۱۳۰ واحد نمونه برداری (برگ) با حداکثر خطای ۴/۳۱ درصد به عنوان اندازه مناسب نمونه انتخاب شد. برای تعیین الگوی توزیع فضایی این کنه از شاخص پراکندگی، روش رگرسیونی (تیلور و آیوانو)، شاخص مورسیتا و شاخص لوید استفاده شد. میانگین تراکم جمعیت، ۲/۰۷۹ کنه به ازای هر برگ بود.

بیشترین جمعیت (۴/۹۳۸ کنه به ازای هر برگ) در ۱۹ مرداد ظاهر شد. شاخص پراکندگی (۵۱۶/۶  $S^2/m=$ )، شاخص تجمع (۲/۶۵۳  $1/k=$ ) و شاخص لوید (۳/۶۵۳  $m^*/m=$ ) پراکندگی تجمعی را برای گونه فوق نشان دادند. روش‌های رگرسیونی تیلور و آیوائو نیز موید تجمعی بودن الگوی توزیع فضایی این گونه بودند. بعضی تغییرات در الگوی پراکندگی از تجمعی به تصادفی در چند تاریخ با استفاده از روش مورسیتا مشاهده شد. سه ضریب تیلور، آیوائو و K به منظور بهینه کردن اندازه نمونه مورد نیاز برای مطالعات آینده استفاده شدند که کوچکترین اندازه نمونه بهینه (۳۴/۷۰) با استفاده از ضرایب تیلور به دست آمد. تعیین پارامترهای الگوی توزیع فضایی *E. frosti* می‌تواند در تخمین تراکم جمعیت این آفت، بهتر شدن برنامه‌های نمونه برداری و مدیریت تلفیقی آن در باغ‌های میوه استفاده شود.